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Uptake Prediction of Eight Potentially Toxic Elements by *Pistia stratiotes* L. Grown in the Al-Sero Drain (South Nile Delta, Egypt): A Biomonitoring Approach

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Abstract: The potential to utilise the free-floating macrophyte *Pistia stratiotes* L. to survey contamination of the Al-Sero Drain in the South Nile Delta, Egypt, by eight potentially toxic elements (PTEs) was investigated in this study. This study considered the absorption of eight PTEs (Cd, Co, Cu, Fe, Mn, Ni, Pb, and Zn), and the evaluated *P. stratiotes* were located in three sampling locations along the Al-Sero Drain, with sampling conducted in both monospecific and homogenous *P. stratiotes*. Samples of both types of *P. stratiotes* and water were collected on a monthly basis between May 2013 and April 2014 at each location, utilising three randomly chosen 0.5 × 0.5 m quadrats. Regression models were designed to predict the concentration of the PTEs within the plant's shoot and root systems. Elevated water Fe levels were correlated with a rise in shoot system Fe concentration, whereas higher Ni concentrations in the water led to a higher Ni concentration within the root system. The latter was also true for Pb. Water Cu levels had a negative association with the Cu concentration within the *P. stratiotes* shoot system. Raised Fe levels were also correlated with a diminished Fe level within the roots. For all PTEs, *P. stratiotes* was characterised by a bioconcentration factor of more than 1.0, and for the majority by a translocation factor of less than 1.0. The goodness of fit for most of the designed models, as indicated by high R^2 values and low mean averaged errors, demonstrated the associations between actual and predicted PTE concentrations. Any disparity between measured and predicted parameters failed to reach significance with Student *t*-tests, reinforcing the predictive abilities of the designed models. Thus, these novel models have potential value for the prediction of PTE uptake by *P. stratiotes* macrophytes inhabiting the Al-Sero Drain. Furthermore, the macrophyte's constituents indicate the long-term impact of water contamination; this supports the potential future use of *P. stratiotes* for biomonitoring the majority of the PTEs evaluated in this study.

Keywords: bioaccumulation and translocation factors; drains; macrophytes; phytoremediation; regression models; water lettuce

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1. Introduction

From the time of the industrial revolution, environmental pollution from potentially toxic elements (PTEs) has been increasing, with grave ecological consequences [1]. Globally, the effects of this pollution on the environment have generated a perilous situation due to the continued accelerated advancement of industrial endeavours [2]. Aquatic ecosystems are particularly at risk of contamination by PTEs. Their pollution is a major issue, since PTEs are persistent in the environment and become biomagnified as they pass through the

food chain [3]. Not only are PTEs toxic, but they accumulate within the environment and are not biodegradable. They therefore represent a serious threat to aquatic ecosystems and to humans [4]. Their transfer through the food chain leads to their transference and accrual within animal organisms, including humans, damaging genetic material and leading to mutations and tumourigenesis [5,6].

Although PTEs are intrinsically present within the environment, commercial, artificial, and agronomic enterprises have all contributed to increased PTE levels [7]. PTEs are predominantly liberated into aqueous bodies from a number of activities, e.g., mining, urban wastewater, smelters, tanning industries, and the textile and chemical industries [8]. Thus, it is important to elucidate the mechanisms underlying transference of PTEs between water/soil and plants. Various models have been designed to understand these processes [9–13]. Furthermore, an appraisal of the amounts of PTEs taken up by vegetation is essential in order to quantify the hazard and for the purpose of environmental governance [14].

Drains are a man-made water collection system; excess surface water from nearby arable land and roads empties into these systems [15]. Drainage systems have been extended and adapted to suit different land use needs. These systems also act as channels for the removal of unsanitised human waste in areas without treatment plants [16]. Mitigating water pollution through the use of vegetated drains is generating considerable attention as an alternative to traditional water treatment processing [17]. Water treatment mechanisms utilising plants are cheap and are attractive to developing nations for wastewater recycling, particularly where PTEs are an issue [18]. The total areas of the Nile Delta and the Nile Valley are estimated to be 22,000 and 13,000 km^2, respectively. The latter therefore forms nearly 63% of the agrarian area within Egypt [19]. The majority of worked Egyptian territories are irrigated via a mesh of channels that coalesce with a parallel connecting system of draining conduits [20]. The sum of the lengths of the channels from the two systems is over 47,000 km; irrigation canals comprise 31,000 km [21].

Pistia stratiotes L. (*Araceae*), a floating macrophyte, also referred to as water lettuce, proliferates vegetatively [22] and is in fact registered as an invasive species in the Global Invasive Species Database [23]. It is widely found within tropical and subtropical areas, but not in Antarctica [24]. Within Egypt, *P. stratiotes* can be observed within the slow-moving canals in the northern Nile Delta territory; in Embaba and in proximity to Cairo [25]; and in static and tranquil waters, particularly in the Fariskur area [22]. It has also been observed at a number of sites in Lake Mariut [26] and Lake Manzala [27] in the northern area of the Nile Delta. *P. stratiotes* is an invasive macrophyte that propagates rampantly, to the detriment of other vegetation such as *Eichorrnia crassipes* (C. Mart.) Solms and *Lemna gibba* L., within the drainage system in the Nile Delta [28]. It has been demonstrated that this species may play a key role in influencing water quality, as it has the capacity to uptake PTEs from wastewater [26,28].

A straightforward appraisal of the transfer factors involved could provide an approximate gauge of the spectrum of PTE transfer, although such as assessment would fail to appreciate precise site-specific properties [29]. However, regression models are mathematical strategies that could anticipate PTE concentrations in macrophyte vegetation by considering variables pertaining to water or soil, e.g., PTE concentration and pH [10–13]. They therefore represent a valuable tool for the assessment of PTE concentrations within macrophytes. Although *P. stratiotes* has been the subject of considerable recent phytoremediation research [28,30–37], there is a dearth of published prediction models for PTE uptake within the shoot and root systems of *P. stratiotes* growing in natural environments. Mathematical models describing PTE uptake by *P. stratiotes* grown on paper mill effluent in a lab scale phytoremediation experiment were developed by Kumar et al. [10]; however, these models cannot be used to predict PTE uptake in conditions other than those used in the experiment. Thus, the aim of the current research was to design a de novo regression model to predict PTE concentration within *P. stratiotes* shoot and root systems in a natural habitat (the Al-Sero Drain), considering water characteristics such as the PTE concentration

and pH. Another goal was to discover how capable *P. stratiotes* could be as a biomonitor of eight PTE concentrations in the Al-Sero Drain, a site considered typical of the South Nile Delta drainage channels. Our hypothesis was that the PTE accumulation capabilities of *P. stratiotes* and its potential to serve as a biomonitor for PTE contamination could differ among populations grown under natural conditions and those grown under experimental conditions. This work will additionally be of value for the future utilisation of this form of vegetation in Egyptian phytoremediation research.

2. Materials and Methods

2.1. Study Area

The research location was in Giza Province, within the Egyptian South Nile Delta region (Figure 1). This territory is classified as hyperarid [38]. The yearly average climate parameters include precipitation in the region of 87 mm, maximum temperature of 30.0 °C and minimum of 14.8 °C, evaporation rate of 6.9 mm/day (Piche), relative humidity of 45.5%, and wind speed of 3.9 m/s [39].



Figure 1. Satellite images of the study area, indicating the locations of the three sampling sites ( ).

2.2. Field and Laboratory

Three sampling locations were selected in relation to the Al-Sero Drain, which comprised monospecific and homogeneous stands of *P. stratiotes* (Figure 1). The site coordinates were (i) site 1: Lat. 30°03′18.88″ N, Long. 31°08′17.56″ E; (ii) site 2: Lat. 30°03′15.73″ N, Long. 31°08′28.20″ E; and (iii) site 3: Lat. 30°03′30.00″ N, Long. 31°08′14.00″ E. *P. stratiotes* biomass was sampled on a monthly basis between May 2013 and April 2014 at each site, utilising three randomly chosen 0.5 × 0.5 m quadrats. The entire population of *P. stratiotes* from each quadrat was harvested, stored in plastic bags, and then transported to the laboratory. The total biomass ranged between 29.9 g DM/m^2 in May and 341.6 g DM/m^2 in August. Detailed data on the biomass were presented in our previous paper [40].

The samples were divided into shoot and root systems and washed with tap water, and then cleaned with deionised water over a 4 mm mesh sieve to eliminate PTEs adsorbed

on the tissue surface and to minimise material loss. In this way, only PTEs absorbed by the plant were determined, and then the bioaccumulation was assessed. The plant material was then reduced to a uniform mass by oven-drying at a temperature of 85 °C. A metal-free plastic mill (Philips HR2221/01, Philips, Shanghai, China) was used to pulverise the dried plant systems, which were then transferred and stored in a desiccator in sterile Ziploc bags. One composite sample from each quadrat from each *P. stratiotes* shoot and root systems at each of the three sampling sites per month was then utilised to assay cadmium (Cd), cobalt (Co), copper (Cu), iron (Fe), manganese (Mn), nickel (Ni), lead (Pb), and zinc (Zn) levels. In total, 108 plant samples per each *P. stratiotes* shoot and root system (3 quadrats × 3 sampling locations × 12 sampling times (months)) were used to determine the uptake of the eight PTEs.

2.3. Water Sampling

Although the water PTE concentrations have not varied significantly in recent years [41], throughout this study, monthly samples were taken over a period of 12 months (May 2013–April 2014), which should have captured the variations in concentration in different months. Three water samples were gathered each month from the same sampling quadrats at each location. The samples were collected, utilising plastic bottles rinsed with deionised water, as coalesced composite samples from the water surface to a depth of 50 cm. At the laboratory, filtration was performed with Whatman membrane nylon filters (pore size 0.45 µm, diameter 47 mm), and then the samples were frozen at −20 °C, pending subsequent PTE analysis of Cd, Co, Cu, Fe, Mn, Ni, Pb, and Zn. This process has been detailed by the American Public Health Association [42].

2.4. Chemical Analysis

The eight PTEs under examination were subtracted from 0.5–1 g of the macrophyte's shoot and root tissues by deploying a mixed-acid digestion technique, using $HNO_3/HClO_4/HF$, 1:1:2, $v/v/v$, in a microwave sample preparation system (PerkinElmer Titan MPS, PerkinElmer Inc., Waltham, Massachusetts, USA). The process was continued until the mixture lost its opacity. The plant digests were then filtered, and double deionised water was used to dilute the samples to 25 mL. Inductively coupled plasma optical emission spectrometry (ICP-OES) (Thermo Scientific iCAP 7000 Plus Series; Thermo Fisher Scientific, Waltham, MA, USA) was utilised for both *P. stratiotes* and the water samples in order to measure the PTE concentrations. Concentrations were given on the basis of dried matter, and deionised water was utilised at all times. Washed glassware and analytical grade reagents were employed appropriately. Instrument readouts were rectified utilising blank reagents. Standard solutions with established concentrations of Cd, Co, Cu, Fe, Mn, Ni, Pb, and Zn were used to calibrate the system. The instrument parameters and operating circumstances were set in keeping with the vendor's operational guidelines. The PTE detection limits were Fe, Pb and Zn, 5.0 µg/L; Ni, 3.0 µg/L; Co and Cu, 0.5 µg/L; Mn, 0.3 µg/L; and Cd, 0.1 µg/L.

2.5. Quality Assurance and Quality Control

With the use of a certified reference material, SRM 1573a (tomato leaves), we confirmed the precision of the PTE test system. The reference material was digested and underwent the same analytical process as the shoot and root systems from the *P. stratiotes* samples on three replicates. The assayed concentrations were contrasted with the certified parameters, and then the percentage was calculated as an expression of accuracy. The spectrum of recovery rates was 96.5–104.3%.

2.6. Data Analysis

Student's *t*-tests were used to analyse any variations in the PTE data between the shoot and root samples. The bioconcentration factor (BCF) was computed in order to establish the efficacy of PTE uptake from the water by *P. stratiotes*, where [43]

$$\text{BCF} = (\text{PTE concentration (mg/kg) in the root system})/(\text{PTE concentration (mg/L) in the water from the same site}) \quad (1)$$

In order to assess the capacity of *P. stratiotes* to transport a particular PTE from its root to shoot system, we calculated the translocation factor (TF) [43]:

$$\text{TF} = (\text{PTE concentration (mg/kg) in the shoot system})/(\text{PTE concentration (mg/kg) in the root system}) \quad (2)$$

Prior to conducting a one-way analysis of variance (ANOVA-1), we evaluated the BCF and TF data by using the Shapiro–Wilk *W* and Levene tests for the presence of a normal distribution and variance homogeneity. The data were then transformed into logs if necessary. An ANOVA-1 was performed on the BCF and TF results in order to identify any variation between the eight PTEs. Any significant variations between the means were established using Tukey's HSD test at $p < 0.05$.

Water pH and its PTE concentration are the principal variables governing the PTE concentration in *P. stratiotes* [10]. The model's general equation can be expressed as [10]

$$C_{plant} = a + (b \times C_{water}) + (c \times \text{pH}) \quad (3)$$

where C_{plant} and C_{water} represent a given PTE's concentration in *P. stratiotes* tissue and water, respectively, and *a*, *b*, and *c* pertain to the regression coefficients.

There was little variation within the results from the three selected sampling areas (data not presented). In view of this, monthly gathered data from two of the sites ($n = 72$) were employed to establish the regression equations for the prediction of the PTE concentrations within *P. stratiotes* root and shoot tissues on the basis of the water indices of pH and the respective PTE concentrations as independent variables. The results from the remaining sampling location ($n = 36$) were kept as a validation dataset.

The determination coefficient, R^2; model efficiency, *ME*; and model strength were used to appraise the quality of the model. Model strength was based on the mean normalised average error, *MNAE*. These parameters were computed according to the equations presented below [44]:

$$ME = 1 - \{\sum (C_{\text{model}} - C_{\text{measured}})^2 / \sum (C_{\text{measured}} - C_{\text{mean}})^2\} \quad (4)$$

$$MNAE = \{\sum (C_{\text{model}} - C_{\text{measured}})/(C_{\text{measured}})\}/n \quad (5)$$

where C_{model}, C_{measured}, and C_{mean} represent the model-predicted, measured, and mean of the measured concentrations of a given PTE, respectively, and *n* is the observation number.

The resulting regression equations were used to estimate the PTE concentrations of the validation. The deviations between the estimated and measured PTE concentrations relating to the same tissue were analysed utilising a Student's *t*-test. The correlation between the PTE levels in the water and the BCF of the PTEs in the *P. stratiotes* root system was measured using non-linear regression. Statistica software, version 7.0 [45], was utilised for all data analysis.

3. Results

Chemical analysis of the water samples taken from the three locations along the Al-Sero Drain revealed a modestly alkaline water, with a mean pH of 7.5 (Table 1). The spectrum of PTE concentrations varied from Cd, 3.5 µg/L to Fe, 523.6 µg/L. The concentration level from high to low of each material was Fe > Pb > Mn > Ni > Zn > Co > Cu > Cd.

Differences in concentrations of six of the PTEs, i.e., not Cd and Pb, between *P. stratiotes* shoot and root systems, were significant (Table 2). Furthermore, the majority of the PTEs were found in higher concentrations in the root system, as opposed to in the shoots. Within *P. stratiotes*, the mean PTE concentration ranges were as follows: Cd, 0.9–1.0 mg/kg; Co, 5.2–17.6 mg/kg; Cu, 10.0–55.5 mg/kg; Fe, 974.1–2511.0 mg/kg; Mn, 331.5–1160.7 mg/kg; Ni, 6.8–20.4 mg/kg; Pb, 39.8–42.0 mg/kg; and Zn, 37.1–48.2 mg/kg. The decreasing orders of PTE concentrations within the shoot and root systems were Fe > Mn > Cu > Pb > Zn > Ni > Co > Cd and Fe > Mn > Zn > Pb > Ni > Co > Cu > Cd, respectively.

Table 1. Potentially toxic element (PTE) concentrations and pH of the water from three sites in the Al-Sero Drain (South Nile Delta, Egypt), supporting the growth of *Pistia stratiotes* populations for one year (May 2013–April 2014).

Value	pH	PTE Concentration (µg/L)							
		Cd	Co	Cu	Fe	Mn	Ni	Pb	Zn
Minimum	7.0	1.0	3.0	1.0	62.0	4.0	7.0	243.0	9.0
Maximum	8.9	36.0	100.0	22.0	980.0	1160.0	110.0	461.0	200.0
Mean (n = 108)	7.5	3.5	20.0	8.2	523.6	234.7	47.3	308.2	26.5
CV (%)	5.7	120.9	70.7	75.4	52.3	112.9	34.6	9.2	95.1

CV: coefficient of variance.

Table 2. Shoot and root potentially toxic element (PTE) concentrations in *Pistia stratiotes* populations growing in the Al-Sero Drain, Egypt (South Nile Delta), over one year (May 2013–April 2014).

Tissue	Value	PTE Concentration (mg/kg)							
		Cd	Co	Cu	Fe	Mn	Ni	Pb	Zn
Shoot system	Minimum	0.1	0.2	2.0	35.0	105.7	0.1	1.2	18.9
	Maximum	3.5	13.5	200.3	3272.0	852.5	50.6	73.1	113.9
	Mean (n = 108)	0.9	5.2	55.5	974.1	331.5	6.8	42.0	37.1
	CV (%)	87.1	65.7	93.8	89.5	59.3	120.3	41.7	46.5
Root system	Minimum	0.1	0.5	3.3	288.0	200.5	1.1	2.7	20.4
	Maximum	3.4	35.2	93.0	5637.0	1474.4	90.6	60.5	124.7
	Mean (n = 108)	1.0	17.6	10.0	2511.0	1160.7	20.4	39.8	48.2
	CV (%)	66.5	55.8	105.8	63.4	33.5	59.5	42.6	40.9
t-value		0.4 ^{ns}	12.5 ***	8.8 ***	8.4 ***	26.2 ***	11.0 ***	0.9 ^{ns}	4.4 ***
Phytotoxic range ‡		5–30	15–50	20–100	>1000	400–1000	40–246	30–300	100–500

t-values represent Student's t-test. ***: $p < 0.001$, ns: not significant (i.e., $p > 0.05$), CV: coefficient of variance. ‡: Kabata–Pendias [46].

The higher water Fe concentration was correlated with the Fe concentration of the shoots ($r = 0.335$, $p < 0.001$) (Figure 2, Table S1). An elevated concentration of Ni in the water was related to the root system's Ni concentration ($r = 0.212$, $p < 0.05$). Elevated water and root system Pb quantities were also associated with each other ($r = 0.294$, $p < 0.01$). The water Cu concentration was negatively related to the Cu concentration within the shoot system ($r = -0.589$, $p < 0.001$). The elevated Fe concentration in the water was associated with reduced Fe in the roots ($r = -0.287$, $p < 0.01$).

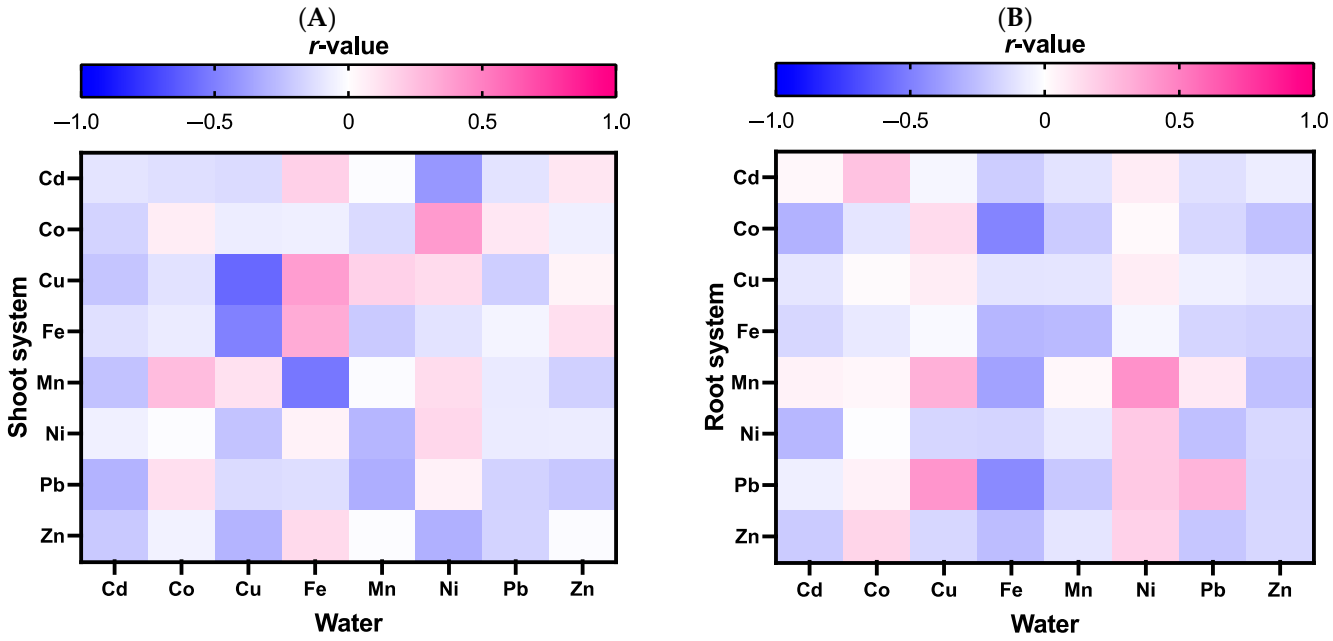


Figure 2. The Pearson coefficient of correlation (*r*-values, $n = 108$) of potentially toxic elements in *Pistia stratiotes* over one year ((**A**): shoot system, (**B**): root system) and their concentration in the Al-Sero Drain waters (South Nile Delta, Egypt) (May 2013–April 2014).

A BCF > 1.0 was calculated for *P. stratiotes* for all the PTEs (Table 3). The values of the parameter were diverse, being generally higher for Mn, and then in descending order: Fe > Cu > Zn > Co > Cd > Ni > Pb. In this study, the TF values also differed according to the PTE under study (Table 3). A TF for the majority of the PTEs for *P. stratiotes* was computed to be <1.0. The TF ranking from root system to shoot system was as follows: Cu > Cd > Pb > Zn > Fe > Co > Ni > Mn. Figure 3 depicts the non-linear regression analysis conducted between the water concentration and the *P. stratiotes* BCF for these PTEs. The BCFs were noted to be maximal at lower water PTE concentrations; they demonstrated an exponential fall with rising PTE concentrations in the water. The R^2 of these exponential equations varied from Pb 0.037 to Mn 0.974.

Table 3. Mean ± standard error ($n = 108$) of bioconcentration factors (BCFs), from the water to root system, and translocation factors (TFs), from the root to shoot system, of potentially toxic elements (PTEs) in *Pistia stratiotes* populations grown in the Al-Sero Drain (South Nile Delta, Egypt) over one year (May 2013–April 2014).

PTE	BCF	TF
Cd	520.7 ± 52.3a	2.1 ± 0.4b
Co	1418.3 ± 151.6a	0.6 ± 0.1a
Cu	2990.3 ± 368.3a	7.1 ± 0.8c
Fe	8974.2 ± 1136.9a	0.7 ± 0.1a
Mn	39,642.5 ± 8247.3b	0.3 ± 0.0a
Ni	471.7 ± 24.6a	0.4 ± 0.1a
Pb	128.7 ± 5.2a	1.9 ± 0.3b
Zn	2316.3 ± 119.3a	0.9 ± 0.1ab
F-value	20.9 ***	50.7 ***

F-values represent a one-way ANOVA, degrees of freedom = 7. Means in the same column followed by different letters are significantly different at $p < 0.05$ according to Tukey's HSD test. ***: $p < 0.001$.

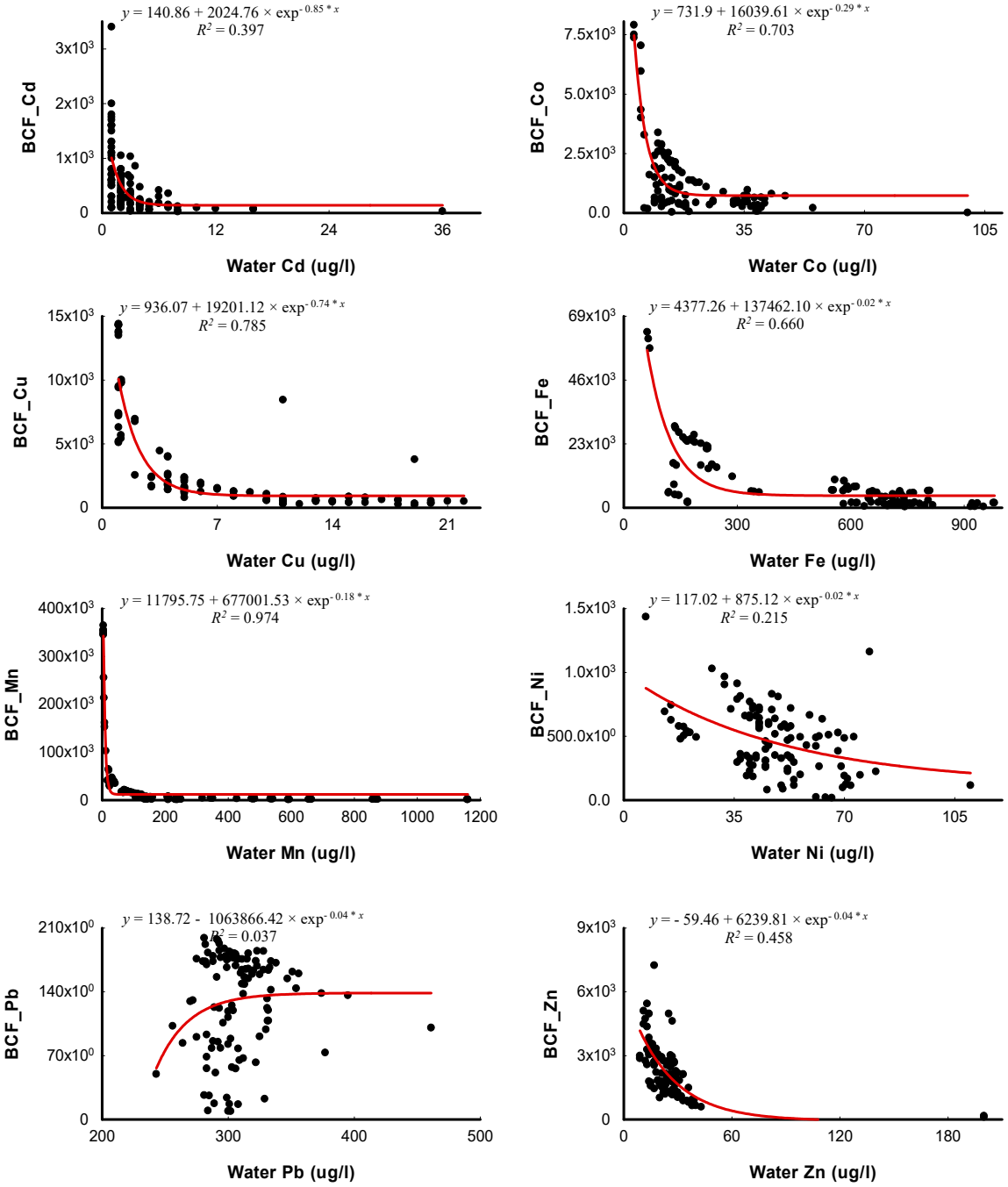


Figure 3. Scatter plot for the bioconcentration factor (BCF) values of eight potentially toxic elements in *Pistia stratiotes* root systems with respect to their concentrations in the water, from three sites in the Al-Sero Drain (South Nile Delta, Egypt), over one year (May 2013–April 2014).

Regression models were designed to predict *P. stratiotes* root and shoot PTE concentrations on the basis of the latter's water concentration and utilising the water pH as a cofactor. Table 4 illustrates the results from these models, as well as their predictive accuracy. Associations between measured and predicted PTE concentrations, together with high R^2 and low mean averaged errors, provided an indication of the acceptability of most of the models. In addition, *t*-test values, which were utilised to analyse any difference between real and predicted concentrations for the eight PTEs in *P. stratiotes* root and shoot systems, were nonsignificant, highlighting the accuracy of the models. For all the models tested, R^2 varied from 0.147 for Cu within the root system to 0.592 for Mn within the shoot system. *ME* parameters had a range between 0.367 for Cu within the root system and 0.811 for Mn within

the shoot system. Furthermore, a low *MNAE* for the majority of the PTEs was observed in relation to the regression models, with a spectrum ranging from 0.179 for Mn within the shoot system to 0.628 for Cu within the root system. With respect to the shoot system, the model for Mn had the greatest R^2 value (0.592) and was related to a high *ME* of 0.811 but a small *MNAE* of 0.179. In relation to the root system, the model for Pb demonstrated the highest R^2 (0.405), with a high *ME* of 0.742 and the smallest *MNAE* of 0.248.

Table 4. Models of regression between potentially toxic elements in *Pistia stratiotes* (mg/kg) and potentially toxic elements in water (µg/L) and pH.

Equation	R^2	*ME*	*MNAE*	Student's *t*-Test	
				t-Value	*p*
Shoot system					
$Cd = 7.74 - 0.14 \times Cd_{water} - 0.83 \times pH$	0.299 ***	0.664	0.336	0.738	0.465
$Co = 29.70 + 0.18 \times Co_{water} - 3.71 \times pH$	0.325 ***	0.716	0.254	0.728	0.471
$Cu = 257.25 - 4.77 \times Cu_{water} - 21.58 \times pH$	0.518 ***	0.748	0.215	0.378	0.708
$Fe = 4531.40 + 0.67 \times Fe_{water} - 529.12 \times pH$	0.279 ***	0.605	0.382	0.954	0.347
$Mn = -1863.31 + 0.06 \times Mn_{water} + 289.66 \times pH$	0.592 ***	0.811	0.179	0.121	0.904
$Ni = 20.94 + 0.09 \times Ni_{water} - 2.69 \times pH$	0.225 ***	0.575	0.472	1.361	0.182
$Pb = 113.52 - 0.20 \times Pb_{water} - 1.07 \times pH$	0.186 ***	0.569	0.575	1.433	0.161
$Zn = -28.60 + 0.21 \times Zn_{water} + 8.05 \times pH$	0.157 ***	0.386	0.591	1.800	0.080
Root system					
$Cd = 7.37 - 0.04 \times Cd_{water} - 0.86 \times pH$	0.294 ***	0.625	0.351	0.953	0.346
$Co = -27.23 - 0.17 \times Co_{water} + 6.53 \times pH$	0.253 ***	0.581	0.433	1.329	0.192
$Cu = 3.90 - 0.11 \times Cu_{water} + 0.74 \times pH$	0.147 ***	0.367	0.628	1.969	0.057
$Fe = 8457.90 - 2.44 \times Fe_{water} - 628.29 \times pH$	0.212 ***	0.571	0.512	1.387	0.173
$Mn = -482.67 + 0.16 \times Mn_{water} + 213.23 \times pH$	0.173 ***	0.445	0.585	1.734	0.092
$Ni = 9.66 + 0.23 \times Ni_{water} + 0.16 \times pH$	0.177 ***	0.492	0.584	1.562	0.127
$Pb = -157.31 + 0.29 \times Pb_{water} + 14.22 \times pH$	0.405 ***	0.742	0.248	0.663	0.512
$Zn = -51.83 - 0.08 \times Zn_{water} + 13.50 \times pH$	0.263 ***	0.600	0.431	0.960	0.343

R^2: coefficient of determination, *ME*: model efficiency, *MNAE*: mean normalised average error, ***: $p < 0.001$. The estimated concentration of a potentially toxic element in a tissue was compared to the measured concentration of the same potentially toxic element using Student's *t*-test.

4. Discussion

This study demonstrated that the majority of PTE concentrations were notably elevated in *P. stratiotes* root systems, rather than in the shoot system. Numerous studies have reported similar findings [10,28,30,34,36,47,48]. This large PTE accumulation within the roots is likely to be a consequence of the PTEs forming complexes with sulphydryl residues, resulting in a lower concentration of free PTE to be transported into the shoots [49]. A number of publications have also described phytochelatin production; these compounds have the ability to sequester PTEs, again contributing to the retention of PTEs inside the roots [50]. Another reason for the higher root concentration is that the root system is the initial point of contact with the PTEs contained within the water [51]. The mean Cu and Pb concentrations recorded for the *P. stratiotes* shoot system in this study were within the phytotoxic ranges; the mean Cd, Co, Fe, Mn, Ni, and Zn concentrations were lower than the phytotoxic range [46]. The mean Co and Pb concentrations recorded for the *P. stratiotes* root system were within the phytotoxic ranges; the mean Cd, Cu, Ni, and Zn concentrations were lower than the phytotoxic range; and the mean Fe and Mn concentrations were higher than the phytotoxic range [46].

It has been shown that aquatic macrophytes are key actors in the extraction of PTEs from wastewater [52]. *P. stratiotes* functions in water pollution removal [28,30–34,36,37]; it is a relatively low-cost method, and in itself is environmentally sound [28]. *P. stratiotes* is typically utilised in constructing wetlands in order to improve the quality of water in water treatment systems [35]. Its advantages include its ability to propagate [53], as well as its PTEs assimilation capabilities [28]. Within the root and shoot systems of *P. stratiotes*, Fe, and then Mn, Zn, and Cu, were found in the highest concentrations, reflecting the straight-

forward underlying mechanisms for their uptake, as they are intrinsically necessary for the proliferation of most vegetation [54]. Similar findings were noted by Kumar et al. [10] for current species grown on paper mill effluent in a lab scale phytoremediation experiment, and by Eid et al. [55] for *E. crassipes* grown in irrigation canals in the North Nile Delta in Egypt. Fe is a critical minor nutrient for both vegetative and animal organisms. In the former, it is an essential component of chlorophyll; over 50% of a leaf's Fe content is within the chloroplasts. This element additionally influences photosynthesis and biomass [56]. Fe and Mn are integrated within the complex of the enzyme nitrogenase, which is necessary for nitrogen fixation through symbiotic and non-symbiotic mechanisms [57]. Zn is also mandatory for both plants and animals, as it is related to numerous enzymes and specific proteins [58]. Both Mn and Zn act as part of the link between an enzyme and its substrate; Mn plays a role in nitrogen transformations in many plants and microorganisms.

Plants and animals also require Cu, which is again associated with enzyme function, especially those which trigger oxidative processes utilising molecular oxygen [59]. Cu is also a constituent of the photosynthesis pathway [60]. Despite the presence of high Pb concentrations within *P. stratiotes* samples, Pb per se is not necessary for plants survival but is carried into plants with other elements. Pb is toxic and is not associated with any notable biological function [61]. In contrast, there was a relatively low uptake of Cd into *P. stratiotes*, a result which reflected that of earlier publications [26,28,43]. Cd is extremely poisonous and is effectively a surplus waste substance discarded from metal refining and electroplating industries that contaminates the environment [58]. It impacts vegetative propagation, metabolism, and water status [62]. Furthermore, Cd acts as an inhibitor of enzymes within the chlorophyll biosynthesis pathway and thus decreases plant chlorophyll content [63].

Monitoring systems for evaluating the accumulation and effect of PTE contamination within aquatic ecosystems are often reliant on live organisms [64]. In this study, there were significant associations between the water concentration of several PTEs and the concentrations of these elements within *P. stratiotes* tissues, thus offering a measure of the amassed consequences of PTE pollution in drain water and a means by which to quantify the quality of the environment. This implies that *P. stratiotes* can act as an effective biomonitor of the presence of PTEs. Furthermore, vegetation containing notable concentrations of PTEs are now being viewed as possible measures of the availability of such elements [43]. It was also noted that some of the positive associations of water and *P. stratiotes* PTE concentrations failed to reach significance, implying that the macrophyte's uptake of all the PTEs present was inconsistent. PTE absorption into *P. stratiotes* was therefore not dependent on the water concentration of the PTEs in every instance [65]. Similar data related to the association between the PTE concentration of the water and *P. stratiotes* have been published in previous studies [10,26,28].

PTE distributions within vegetative tissues are not generally uniform in plants from either aquatic or terrestrial ecosystems [26,66]. Their accumulation in various species occurs in accordance with multiple factors, including chemical speciation, water transport, plant species and accompanying phenology, physiology, vigour, propagation and age, climatic parameters, salinity, pH, and interchelating of the PTEs [43,51,67,68]. Calculating the BCF is a straightforward technique to measure the translocation of accessible PTEs from either the soil or water into a plant's root system [69], whereas transport from the root to shoot system can be appraised utilising the TF. Yanqun et al. [70] published data indicating that, in plants, accrued PTEs have a BCF > 1.0, whereas in plants that exclude PTEs, the BCF < 1.0. The current research demonstrated a BCF > 1.0 for *P. stratiotes* in relation to all the PTEs tested, indicating the ability of this macrophyte to absorb PTEs within its root system, as well as its appropriateness for phytoremediation or rhizofiltration tasks. These data essentially mirrored work published by Galal et al. [28] and Kumar et al. [10]. The fact that *P. stratiotes* is recognised as being a possible candidate for phytoremediation reflects the view of Weis and Weis [71], who have also reported that PTEs can be accumulated by aquatic plant species through their root systems. Overall, Mn had the largest BCF, with

lower values in descending order for Fe, Cu, Zn, Co, Cd, Ni, and Pb. Mn, Fe, Cu, and Zn exhibited a higher BCF as they are essential macronutrients for the macrophyte.

In the present study, non-linear regression was used to relate PTEs in *P. stratiotes* root system to the PTEs concentration in the water. The data demonstrated an exponential drop in BCF values for all the PTEs with rising water concentrations of these elements. In other words, the bioaccumulation of PTEs in root system decreased with an increase in PTE concentration in the water. A similar finding was noted by Prasad and Maiti [72] for *E. crassipes* growing in ponds from mining and non-mining areas in India, and Eid et al. [55] for *E. crassipes* grown in irrigation canals in the North Nile Delta in Egypt. A similar inverse relationship was recorded in another investigation in the terrestrial environment by Wang et al. [73] in four common vegetables (Chinese cabbage, spinach, celery, cole) grown on PTE-contaminated soils under field conditions in China. A potential mechanism to explain this is that the plants have a crucial ability to self-regulate PTE uptake into their root systems [74,75]. Additionally, the macrophytes tend to thrive less well in polluted water. This is particularly the case where the water is heavily contaminated; plants undergo blasting and may fail to survive, owing to the poisonous consequences of the water toxins [72]. In this situation, the poor quality of the habitat ameliorates the ability of the macrophytes to absorb PTEs, and thus the concentration of these PTEs within the root system is diminished [73]. The results therefore point to the fact that the concentration of the PTE is important for the availability of PTEs in water.

The TF is a measure of the effectiveness of PTE transfer from the macrophytes' roots to their shoot systems. Calculation of this parameter for *P. stratiotes* revealed some differences between the varying PTEs; the value was < 1.0 for the majority of PTEs evaluated. *P. stratiotes* therefore has the capability to prevent some PTEs from reaching its physiologically active components, e.g., the leaves. The differences seen in the TF values could be associated with the interactions between the PTEs, which can originate from conflicting and synergetic processes [76,77]. Further factors to explain the differences in TF include physiological parameters relating to the plant, PTE solubility and availability factors, and governance pathways within the root and shoot systems which limit translocation to the latter [74,77].

Regression models can be used as mathematical strategies to facilitate the prediction of plant PTE concentrations utilising water parameters, e.g., the PTE concentration and pH [10,11]. Essential related concepts influencing plant absorption include PTE solubility and bioavailability [44]. pH acts as one of the most significant factors to determine the net metal ion availability in aqueous solutions, as well as their further absorption by plants [78]. Thus, the water pH is often involved in such models, as it impacts the bioavailability of the PTEs [10,11]. In the current study, the pH in the Al-Sero Drain ranged between 7.0 and 8.9. In a recently published study, the pH influence on the effectiveness of PTE absorption by the plant was reported as acidic > neutral > basic [29]. A study by Awuah et al. [79] showed that *P. stratiotes* was capable of growing at a minimum optimum pH of 4.4 when grown in ponds for wastewater treatment. Therefore, lowering the pH value of the Al-Sero Drain could enhance plant efficiency of the uptake of all selected PTEs. The results from this study demonstrated the ability of the models to estimate the quantity of PTEs within *P. stratiotes* root and shoot systems, according to parameters of model performance, i.e., R^2, *ME*, *MNAE*, and *t*-values. In the designed models, satisfactory R^2 parameters were calculated in some instances, within a spectrum extending from Cu, 0.147, in the root system, to Mn, 0.592, in the shoot system. The diversity observed indicates that *P. stratiotes* may exhibit some metal-specific uptake properties [10].

The data presented in this study are new, with respect to the generation of regression models, in terms of their use as predictive tools for PTE absorption in *P. stratiotes* grown in a natural environment. To the authors' knowledge, no studies focused on this scenario have been published to date. Thus, the presented data have been contrasted with research conducted within a laboratory setting. Kumar et al. [10] described a range for R^2 for Cd in *P. stratiotes* of 95.0–99.0% when the macrophyte was cultured in paper effluent within a

laboratory sized phytoremediation model. This compares with an R^2 for Cd of 29.4–29.9% measured in this study in a natural habitat. The R^2 for Pb attained by Kumar et al. [10] was between 79.0% and 91.0%; in this study, the range was 18.6–40.5%. The higher values in the former work suggested minimal intersample diversity; the data were collected from macrophytes cultured in a uniform laboratory setting. In the current study, the lower R^2 values may have been due to the fact that the samples were collected over a year, from May 2013 to April 2014, and any diversity in the water conditions and concentrations of PTEs became merged. Additionally, the smaller R^2 parameters in this research may reflect a lack of model sophistication and its restricted ability to demonstrate complex natural PTE phenomena [80].

5. Conclusions

The current research was carried out in order to design new regression models for the prediction of eight PTE concentrations within the root and shoot systems of *P. stratiotes*, from the equivalent water elemental concentrations, utilising the water pH as a cofactor. *P. stratiotes* was characterised by a BCF > 1.0 for all eight PTEs evaluated in the study, and the TF of Cd, Cu, and Pb were > 1.0. This indicates that *P. stratiotes* is suitable for Cd, Cu, and Pb phytoextraction, as well as the exclusion of the remaining PTEs. Moreover, the high BAF and low TF of most investigated PTEs indicate the potential of *P. stratiotes* for phytostabilisation of these PTEs. The majority of the designed models for the prediction of PTE concentrations within the shoot and root systems of this plant were robust, offering a good fit, with high efficacy and minimal error. They could therefore be of use as predictors of PTE accretion within the plant components of *P. stratiotes* that inhabits drainage canals, with the exception of those with a low R^2. These models represent new possibilities for environmental risk assessments and the creation of standards for PTE water quality. An extended field study may be needed for irrigation canals.

Supplementary Materials: The following are available online at https://www.mdpi.com/article/10.3390/su13095276/s1, Table S1: Pearson correlation coefficient (*r*-values, *n* = 108) between potentially toxic elements (PTEs) in *Pistia stratiotes* tissues and their concentrations in the water of the Al–Sero Drain (South Nile Delta, Egypt) over one year (May 2013–April 2014).

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